Chitosan Coating to Extend the Storage Life of Tomatoes

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Abstract. The effect of chitosan on respiration, ethylene production, and quality attributes of tomato (*Lycopersicon esculentum* Mill.) fruit stored at 20°C was investigated. Coating the fruit with chitosan solutions reduced the respiration rate and ethylene production, with greater effect at 2% than 1% chitosan. Coating increased the internal CO₂ and decreased the internal O₂ levels of the tomatoes. Chitosan-coated tomatoes were firmer, higher in titratable acidity, less decayed, and exhibited less red pigmentation than the control fruit at the end of storage. Chemical name used: 2-amino-2-deoxy-p-D-glucan (chitosan).

The major postharvest losses of tomatoes are due to fungal infection, physiological disorders, and physical injuries (Grierson and Kader, 1986). The prevalent method of preservation of postharvest produce is the use of moderately low temperatures and high humidity. For commodities such as tomatoes, however, low temperatures induce chilling injury (Morris, 1982). An alternative method is the use of modified atmospheres (low O₂, high CO₂) (Kader et al., 1989). Atmospheric modification can be achieved by plastic film packaging (Kader et al., 1989), and by individual coating of fruit with waxes, sucrose fatty esters, Pro-Long, or *N,O*-carboxymethylchitosan (Banks, 1984, 1985; Elson et al., 1985; Meheriuk and Lau, 1988; Smith and Stow, 1984). With these coatings, however, problems of fermentation due to the depletion of O₂ and failure to ripen properly have been observed. A desirable coating should extend the storage life of fresh fruit without causing anaerobiosis and at the same time reduce decay.

Chitosan, obtained by deacetylation of chitin, might be an ideal preservative coating for fresh fruit because of its film-forming and biochemical properties (Muzzarelli, 1986). Unlike other coating materials, chitosan is known to be antifungal to several fungi, including *Botrytis cinerea* Pers. ex Fr. and *Rhizopus stolonifer* Ehr ex Fr. (El Ghaouth et al., 1992a, 1992b; Hirano and Nagao, 1989; Kendra and Hadwiger, 1984; Stössel and Leuba, 1984), to induce chitinase, a defense enzyme (Mauch et al., 1984), and to elicit phytoalexin (pisatin) accumu-

lation in pea (*Pisum sativum* L.) pods (Hadwiger and Beckman, 1980; Kendra and Hadwiger, 1984). Furthermore, chitosan, a by-product from the seafood industry, appears to be a safe material as indicated by toxicological studies (Hirano et al., 1990). Since chitosan can form a semipermeable film (Bai et al., 1988), coating fruit with chitosan may modify the internal atmosphere of the tissue and consequently delay ripening. This paper reports the effect of chitosan coating on the storage life of tomato fruit held at 20°C.

Crab-shell chitosan was purchased from ICN, Cleveland, and ground to a fine powder. The chitosan solutions were prepared by dissolving chitosan [1% or 2% (w/v)] in 0.25 N HCl. The solutions were centrifuged to remove undissolved particles, and the pH was adjusted to 5.6 with 1 N NaOH. At this pH, chitosan (pK_a = 6.2) is positively charged and exhibits maximal biological activity (Stössel and Leuba, 1984). Tween 80 [0.1% (v/v)] was added to the solutions to improve wettability.

Mature-green tomato fruit were harvested from the greenhouse at Laval Univ., Quebec, and sorted on the basis of size, color, freedom from blemishes, and fungal infection. Fruit were surface-disinfected by immersion in 10% commercial bleach for 3 min, air-dried, and individually dipped either in 1% (C-1%) or 2% (C-2%) chitosan solution (w/v) or in sterile deionized water (pH 5.6) containing 0.1% (v/v) Tween 80. Each treatment consisted of five replicates of 10 fruit (a total of 50 fruit) in a randomized complete-block design. After air drying, fruit were stored at 20°C in plastic containers (10 fruit per container) with a continuous flow of humidified air [95% relative humidity (RH)].

The respiration rate and ethylene production were determined by sealing individual fruit in 1.35-liter containers for 1 to 3 h, at which time two 5-ml samples of headspace gas were withdrawn with gas-tight hypodermic syringes and analyzed for ethylene and CO₂ using gas chromatography (Model 8500,

Perkin-Elmer, Beaconsfield, Bucks, U.K.). For each treatment, four single-fruit replicates were analyzed. To assess the effect of coating on the internal atmosphere, 1-ml samples of internal gas were withdrawn from the fruit on day 2, 7, and 12 of storage using the method of Banks (Banks, 1984, 1985) and analyzed for CO₂ and O₂. Gas samples were drawn from seven to 12 fruit per treatment.

Tomato fruit were graded visually every 2 days according to the U.S. Dept. of Agriculture Visual Color Chart (U.S. Dept. of Agriculture, 1975), with 1 = mature green; 2 = breaker; 3 = turning or one-third pink; 4 = two-thirds pink; 5 = light red; 6 = red. A total of 40 fruit per treatment was used for color evaluation, and individual fruit scores were averaged to obtain the color index. The incidence of fungal infection was monitored throughout the storage period. Spoiled fruit were discarded.

At the end of 4 weeks of storage, four replicates of eight fruit (a total of 32 fruit) per treatment with complete randomization were used for determination of firmness and titratable acidity. Fruit were cut through the equatorial plane, and the penetration force (N) of the exocarp and endocarp was measured by the method of Holt (Holt, 1970)

Fig. 1. Carbon dioxide evolution (A), ethylene production (B), and changes in ripening index (1 = mature green, 6 = red) (C) of noncoated (□) and 1% (○) and 2% (●) chitosan-coated tomatoes stored at 20°C. Data presented are means of four replicates. The vertical bar in each panel represents the SEM.

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Fig. 2. Internal O₂ (A) and internal CO₂ (B) levels of noncoated (□) and 1% (○) and 2% (●) chitosan-coated tomatoes stored at 20C. Vertical bar represents the SEM.

Table 1. Effect of chitosan coating on quality attributes of tomatoes after 28 days of storage at 20C.

Chitosan concn (%)	Infection (%)	Fruit firmness ^a	Titrateable acidity (mg)
0	28.1 a	2.6 a [*]	106 a
1	9.4 b	3.0 ab	129 b
2	3.1 c	3.3 b	130 b

^aFor firmness and titrateable acidity determination, only fruit at the red-ripe stage were used. The acidity was expressed as milligrams of citric acid per 100 ml of homogenate.

^{*}Means separation by LSD method ($P \leq 0.05$).

with an Instron Universal testing machine (Instron Corp., Canton, Mass.) equipped with a 4-mm flat plunger. Titrateable acidity was determined using a 10-g aliquot of whole-tomato puree and titrating with 0.1 N NaOH to pH 8.1.

All experiments were conducted twice. Data from each experiment were tested by analysis of variance, then means were separated by the protected LSD ($P \leq 0.05$) method.

During storage at 20C, the respiration rate of tomatoes increased gradually (Fig. 1A). Between 2 and 7 days following the mature-green stage (day 0), chitosan-coated fruit had a slightly higher respiration rate than the control. After 10 days of storage, control fruit exhibited a significantly higher respiration rate ($P \leq 0.05$) than the treated fruit. There was no difference in respiration rate between 1% and 2% chitosan-coated fruit until 15 to 19 days of storage, when the respiration rate of C-2% fruit was markedly lower than that of C-1% fruit. Ethylene production followed a climacteric pattern during the storage of all fruit; however, the climacteric trend was suppressed in chitosan-coated tomatoes (Fig. 1B). Fruit coated with chitosan had a significantly lower rate of ethylene

production ($P \leq 0.05$) than the control fruit. No significant difference in ethylene production between C-1% and C-2% fruit was observed until after 13 days of storage. Thereafter, the C-2% fruit had a lower ethylene production than C-1% fruit. Reduction of the respiration rate and ethylene production as a result of coating with edible films has also been reported for banana (*Musa × paradisiaca* L.) (Banks, 1984), apple (*Malus domestica* Borkh.), pear (*Pyrus communis* L.) (Elson et al., 1985; Davis et al., 1989; Meheriuk and Lau, 1988), and tomato fruit (Nisperos and Baldwin, 1988). For tomatoes, however, development of a blotchy appearance and an increase in stem rot were observed with Durkex-500 coating (Nisperos and Baldwin, 1988).

Chitosan also affected fruit color (Fig. 1C). After 22 days of storage at 20C, the color index of the control, C-1%, and C-2% fruit were 5.6 (red), 4.3 (two-thirds pink), and 3.3 (turning), respectively. Tomatoes coated with 2% chitosan became red more slowly than the C-1% fruit.

Chitosan coating decreased the O₂ and raised the CO₂ levels within tomato fruit stored at 20C, with a greater effect at the higher coating concentration (Fig. 2). After 12 days of storage, the internal CO₂ content of C-2% fruit was 160% higher than that of the control, while the O₂ level was 30% lower. The internal O₂ level (12%) resulting from chitosan coating likely would not cause anaerobiosis, which occurs in tomato only at external O₂ levels below 3% (Buescher, 1979).

Chitosan coating reduced the incidence of decay, mainly that caused by *Botrytis cinerea*. In the control fruit, the first signs of fungal development appeared after 2 days of storage, while the signs of infection were visible only after 10 days of storage at 20C in coated fruit. By the end of 4 weeks of storage, the percentage of decayed coated fruit was significantly ($P \leq 0.05$) lower than that of the control (Table 1). C-2% tomato fruit were significantly firmer ($P \leq 0.05$), and both groups were higher in titrateable acidity than the noncoated fruit after 4 weeks at 20C (Table 1). Firmness and the concentration of titrateable acidity were similar for C-1% and C-2% fruit.

The reduction of C₂H₄ and CO₂ production and the retention of firmness and more rapid color development as a result of coating is generally explained by the modification of the endogenous levels of O₂, CO₂, and C₂H₄. Banks (1984) reported that coating banana with Pro Long delayed ripening by lowering the internal O₂ concentration (2% to 6%). In a study with Cox's orange apples, however, Smith and Stow (1984) suggested that the effect of Pro Long on color and firmness was not due solely to the modification of internal CO₂ (8%), because 8% CO₂ during storage did not duplicate the same effect observed with Pro Long coating. Our results do not provide evidence as to whether the observed changes in ripening are mainly due to an increase in internal CO₂ resulting from the differential permeability of the coating. Nonetheless, this possibility should not be

disregarded, especially since high CO₂ (5% and 10% with 21% O₂) has been shown to reduce ethylene production and delay red pigment development in tomatoes (Buescher, 1979), while storage at 3% to 5% O₂ with 0% CO₂ was considered optimal for delaying ripening of tomato (Kader, 1986).

In conclusion, this study demonstrates the potential of using chitosan-derived coatings to delay ripening and reduce decay incidence in tomato fruit. Extensive postharvest storage tests are necessary to determine the feasibility of using these coatings on a commercial scale.

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